We have also applied these proteomic methods to studying protein modification in acute hypoxia. In endothelial cells, there is an increase in cysteine oxidation in several proteins that can mediate acute responses to hypoxia prior to the activation of the HIF pathway, and we are currently studying in more detail the role of protein S-nitrosylation.

We have also recently shown that acute hypoxia produces a superoxide burst in cells, which can be converted in an oxidative signal through protein cysteine modification, and we are unraveling the molecular mechanisms producing this superoxide burst in mitochondria.

http://dx.doi.org/10.1016/j.redox.2015.09.027

Macrophage Polarization In The Tumor Microenvironment

Bernhard Brüne, Andreas Weigert, Nathalie Dehne

Institute of Biochemistry I, Faculty of Medicine, Goethe-University Frankfurt, Frankfurt, Germany

Background: Tumor associated macrophages (TAMs) are known to support tumor progression and their accumulation is generally associated with poor prognosis. The shift from a tumor-attacking to a tumor-supportive macrophage phenotype is based on an educational program that, at least in part, is initiated by apoptotic tumor cells.
Aims: We explored the macrophage phenotype shift during tumor progression by analyzing the macrophage NO-output system and examining potential NO targets.
Methods: Biochemical and Molecular Biology-orientated cell culture experiments, in part using 3D-tumor spheroid models as well as animal experiments were used.
Results: Apoptotic cells polarize macrophages towards a healing, tumor-supportive phenotype. Soluble mediators released from apoptotic cells, among them the lipid sphingosine-1-phosphate (S1P), cause expression of arginase 2 in macrophages, thereby lowering citrulline/NO formation but enhancing ornithine production. Mechanistically, this is achieved via the S1P2 receptor and the CRE (cAMP-response element) binding site in the arginase 2 promoter. Reduced NO-formation is also seen in ex vivo macrophages from a xenograft model allowing restricted vs. unrestricted tumor growth based on tumor-associated S1P-formation. The theoretical ability of NO to target hypoxia-inducible factor-1 (HIF-1) and jumonji histone demethylases (JHDMs) in cells of the tumor microenvironment will be discussed in light of the iNOS/arginase balance. Moreover, data on the importance of HIF-1 in macrophages for their interaction with tumor cells, polarization, and angiogenic potential will be presented.
Conclusions: We hypothesize that apoptotic death of tumor cells and associated macrophage activation facilitates the progression of malignant disease. The macrophage polarization program affects the NO-output system and the capacity of macrophages to support or restrict tumor growth.

http://dx.doi.org/10.1016/j.redox.2015.09.028

Young Investigation Session Selected Oral Communications

Polymorphisms In The Nitric-Oxide Synthase 2 Gene And Prostate Cancer Pathogenesis

Charlotta Ryk, Petra de Verdier, Emmie Montgomery, N. Peter Wiklund, Fredrik Wiklund, Henrik Grönberg

Department of Molecular Medicine and Surgery, Karolinska Institutet, Stockholm, Sweden

Background: Nitric-oxide synthase (NOS)-polymorphisms influence the cellular amount of NO, and are associated with disease-risk in many disorders. We investigated 145 SNP-polymorphisms and a (CCTTT)n-microsatellite in the *NOS2*-gene in 3161 prostate-cancer patients and 2149 controls from a Swedish population-based GWAS-study.
Aim: To analyze possible associations between *NOS2*-polymorphisms, prostate cancer, and prostate cancer pathogenesis.
Methods: Two groups were analyzed, those with advanced tumours (Gleason≥6), and those with tumours of mixed Gleason-statues. Affymetrix 5.0-chip (SNP-polymorphisms), DNA Fragment-analysis and Sequencing ((CCTTT)n-microsatellite) were used for genotyping. Genotypes were combined with information on tumour stage, Gleason, PSA, metastases and cancer-specific death, using clinical follow-up.
Results: We divided the (CCTTT)n-alleles into short (S, $n \leq 10$), intermediate (M, $n = 11$-12) and long (*L*, $n \geq 13$). Patients homozygous for longer repeats (LL) had decreased risk of highly aggressive (Gleason ≥7;PSA > 20;T3+) tumours (OR:0.40;CI:0.14-1.08;p=0.071), but, once ill they showed a threefold increased risk of dying in prostate cancer (HR:3.31;CI:0.85-12.85;$p = 0.084$), compared to SS-homozygotes. The SNP-alleles that co-varied with the (CCTTT)L-allele also had lower risk of aggressive tumours, as well as, once ill, a 2-4 times higher risk of dying ($p = 0.009$). Also the proportion of patients with lymph node metastases increased with length of the (CCTTT)n-alleles of the patients (SS < SM < ∩ SL < MM < ML < LL)(trend analysis; $p = 0.033$).
Conclusions: Nitric oxide can induce proliferation as well as apoptosis depending on cellular context. Our results suggest that *NOS2* polymorphisms may influence the risk of aggressive prostate cancer and that these polymorphisms could have an impact on disease pathogenesis, possibly by affecting intracellular nitric oxide levels.

http://dx.doi.org/10.1016/j.redox.2015.09.029